## HERZSTEIN LECTURES IN THE UNIVERSITY OF CALIFORNIA FOR 1905.

### Special Chemical Problems Related to Practical Medicine.

Synopsis furnished to the JOURNAL by the lecturer.

### LECTURE III.

### The Relation of the Nitrogenous and the Carbonous Metabolism in Disease.

#### By ALONZO ENGLEBERT TAYLOR.

The nitrogen metabolism (including the purin metabolism) may be defined as the metabolism of cellular regeneration. There is in the body cells a constant autolysis. New nuclear substance and protoplasm must be formed for new cells. Beyond the term of growth, the magnitude of these regenerations is quite independent of the activity of the life of the individual, and is related to the species. Apart from conditions of disease, a cell seems to last a certain length of time in each species. The velocity of autolysis and regeneration is very low in the cold-blooded animal and is apparently little less during hibernation than during the active state. If the diet of an animal be properly arranged, it will be found that severe and prolonged exertion is supported entirely by the carbonous metabolism; the nitrogenous metabolism is not increased. Heat is, of course, produced in the nitrogenous metabolism, but it is incidental and comprehends but a fraction of the heat production.

Beyond the metabolism of the cellular life, the endogenous metabolism, there is an exogenous nitrogenous metabolism, related to the excess of protein input over the cellular needs. The limitations and interrelations of these two have not been defined. There is in the purin metabolism also, an endogenous and an exogenous fraction.

The protein necessary to a normal diet may be set down as about one gramme per kilo weight. With a large body weight, the ratio will be disproportionately reduced, since further increase in weight will be largely fat and skeleton, and fatty tissue has small nitrogen needs. Since a body of 70 kilo will contain about 10-11 kilo of cellular material, one gramme of protein per day maintains the status of some 150-200 grammes of cells (calculated water-free). The nitrogen proportional to this protein ratio will appear in the urine daily in the form of end products of the protein catabolism. This output represents the reaction velocity of these cells.

The 150-200 grammes of cellular material has a uniform water content and represents a substrate concentration in a reacting system. The nitrogen output represents the transformation under constant conditions of work and stands for the constant. A corresponding amount of substrate added regularly to the system (in the form of the protein diet) will maintain the concentration of the substrate. The products of the reaction are removed from the body. We know that the transformation in a chemical system will be uniform in time if the products of the reaction be removed, the temperature held constant and the substrate concentration maintained by the regular addition of that amount of the substrate indicated by the constant. The accelerator for the reaction of hydrolysis may be assumed to be the intracellular proteolytic ferments, and these are assumed to be unvarying, since the transformation on a constant diet is uniform. It is not yet possible to apply the law of mass action to the details of the protein metabolism (for example, the substrate is not strictly the mass of the cells, but some relationship between the mass of the cells and the circulating protein), but in this point of view is represented unquestionably the dynamics of the process.

There is a great power of adaptation in this function. It is possible to cut down the protein input materially; it is possible to compel the body to maintain its heat and energy expenditure entirely from protein. It is possible in adaptable individuals to cut the protein ratio down to some 30 grammes of protein per day for a body of 70 kilo, though disproportionate amounts of carbohydrates will be needed in the diet. When the quotum of protein in the diet is normal, we need that amount of fat or sugar that will yield 25-35 Cal. of heat per kilo per day; when we cut the protein down one-half, we need a ratio of carbohydrate or fat sufficient to yield 40 Cal. per kilo per day. This indicates that when the repair of the cellular mass of the body is to be accomplished with the minimum of new material, these cells do not perform their functions of combustion as economically as in the normal.

When no food is taken except protein, the body will perform all its functions on this one material. A body of 70 kilo will require from 500-600 grammes of protein, ten times the amount needed for the repair of the cellular losses. There is evidence that when compelled to maintain the body heat by the combustion of protein, the cells work at a greater wear and tear than normally, and proportionately more than 1 gramme per kilo weight will be needed for their repair; therefore, somewhat more than the isodynamic amount of protein will be needed if all the functions are to be supported by it. There is also evidence that muscular contraction (work) is not maintained as economically by the combustion of protein as by that of sugar.

When to a mixed diet an excess of protein is added, the body resists an excess of circulating protein, so that all the excess is hydrolysed and the nitrogen output balances the input. Since this evolves heat, less of the carbonous substances of the body will be burned, and as the combustion of carbon is regulated by the heat needs and not by the material available, that will be spared and stored as glycogen and fat. The saving is not strictly isodynamic, since the oxidation processes on a heavy protein diet are excessive.

When an animal is starved, the protein catabolism is reduced to somewhere near the figure given as the normal minimum (½ gramme per kilo). The heat is first supported by the combustion of glycogen; later of fat. So long as the fat is abundant, we have the normal relations of protein and carbonous metabolism; when the fat has sunken appreciably, some body protein begins to be burned for heat, and this ratio increases progressively until at the close, when the fat is low, the temperature is maintained largely by the combustion of cellular protein.

When an animal is placed upon a protein diet, the body fat and carbohydrate retain their status quo. The blood sugar rests at a minimum, the glycogen of the body does not increase, the fat is unchanged, the metabolism of fat and sugar have become quiescent. Just how the protein molecule is burned, following its hydrolysis, is not known; it has been usually assumed that the carbonous groups would pass through the stage of sugar, but the direct combustion is theoretically possible. If the protein diet be excessive, it is all hydrolysed and burned, the carbonous as well as the nitrogenous portion. It has been assumed that under these conditions a part of the carbon of the excessive protein would be retained as fat or glycogen. Careful experimentation has thrown the gravest doubt upon this statement, although in occasional instances of forced experimentation small amounts of carbon are retained. The amount concerned is trivial and cannot be used to force an interpretation of the conditions under normal circumstances. Under physiological circumstances a derivation of carbohydrate from protein is improbable.

The carbonous metabolism maintains the body heat and is the basis of muscular energy. The incidental heat developed in the hydrolysis and oxidation of protein in the essential protein catabolism is not over 300 Calories per day. The two functions of combustion for body heat and muscular energy are in part reciprocal. When heat is produced in association with muscular work, that heat maintains the body temperature, with a consequent saving of other combustion. The functions are only in part reciprocal. All notable muscular exertion is associated with a heat production beyond the amount needed to maintain the body temperature, and the excess is dissipated. Within certain limits the processes for the combustion of carbohydrates and fat for the purposes of the maintenance of the body heat are a reflex function of the external temperature; the more rapidly the body loses heat, the more rapidly the process of combustion will operate, and when the loss is so rapid that the direct chemical processes, aided by the maximum available restriction of heat dissipation, cannot maintain the loss, heat will be furnished by the indirect method through muscular exercise, as by shivering. In the other direction, however, the influence is less strict. If we place a human body in a room with a body temperature, it might be conceived that, apart from the small amount of heat produced in the protein catabolism, the body would exhibit no combustions at all, since no heat is needed. This is not the case. There will be a reduction in the combustion of sugar and fat, but a certain amount of heat will be produced, although it is not needed. Higher temperatures are often accompanied by excessive combustions; thus a man in a Turkish or Russian bath will often actually burn up more carbonous substance while within the bath than under ordinary circumstances.

One gramme of fat yields about 9 Calories of heat, a gramme of sugar about 4.2 Calories. Now, in the body the combustion of sugar is more economical than in the case of fat, so that here sugar has, for the purposes of fuel. half the value of fat. To furnish the body heat. we need some 250 grammes of fat or 500 grammes of sugar. These are in the body practically isodynamic in all proportions. There is, however, a difference in their relations to the protein metabolism: sugar has a greater saving power for protein than fat.

Sugar is burned in the body indirectly. One of the great questions now pending is whether sugar is burned via alcohol as an intermediary product, analogous to the fermentation of sugar. When in excess in the body, sugar is converted into glycogen. When the glycogen storage reaches a certain point, sugar is converted into fat. How the fats are burned in the body we do not know. If they are burned directly, it is of importance to know whether this occurs by the successive splitting off of CH. groups, whether it is preceded by an intra-molecular cleavage or whether a fat is burned by direct oxidation, through oxy-acids. It is also possible that fats are not burned directly at all, but first converted into sugar. The easy formation of fat from sugar makes the reversion into sugar a chemical feasibility. In plants this reversion is a regular physiological act. That these combustions are to be interpreted as fermentations is certain.

There is between the carbonous and protein metabolisms one fundamental difference; their relation to the input. The magnitude of the exogenous catabolism of protein is a function of the input; combustion in the carbonous metabolism is a function of the body needs. The body needs only some 60 grammes of protein per day, but, no matter how much more is ingested, all the excess will be promptly hydrolysed and eliminated. Of fat and sugar the body will burn only so much as is needed for the body heat and for muscular exertion; if more than this amount be ingested, it will not be burned, but stored. The body

invites an excess of carbon; it repels an excess of nitrogen. The body stores the carbon for future heat and work; it will not store the nitrogen for future regeneration of cells. That a small retention of protein can be accomplished is true, but so soon as the diet is returned to the normal, the excess retained will be hydrolysed. This distinction is fundamental, and of the greatest physiological and practical importance. It means chemically that in the case of protein the retained excess adds to the substrate concentration, is active in the sense of the mass law; the body possesses no power to store it in some inactive form. In the case of fat and sugar the excess retained is stored in an inactive form, so that the substrate concentration is not increased. Sugar is the active state of carbohydrate, glycogen is the storage state, a soluble fat-complex is the active state of the fat, neutral fat is the storage state.

*Variations in Conditions of Disease.* The difficulties that attend the reduction of obesity and the fattening of a naturally emaciated individual have given the impression that the fat person subsists on less food and the thin person requires more food than the normal. Assuming that in each instance the factor of digestion and assimilation is normal, are there persons whose body needs are appreciably greater or less than the normal? Is there such a thing as a retardation of metabolism as a type, a constitutional entity that is expressed in a reduction of the unit-metabolism? The bear has a different unit of metabolism when active and hibernating. Such a constitutional retardation as postulated by Bouchard would resemble the hibernating metabolism as against the active metabolism. Is there such a thing as a constitutional type of excessive metabolism? There are here many individual variations. These variations are much less marked for the nitrogenous than for the carbonous metabolism. Apart from disease, the protein needs of the bodies of different individuals display small variations. The individual variations in the carbonous metabolism are normally very great, as is to be expected from the number of adventitious variables. The following factors must be considered: (a) The relation of body-weight to surface dimensions (the dimensions of heat radiation) are different for different individuals. (b) The variations in the physical modes of heat dissipation are different, depending on the pigmentation and the thickness of the skin, the vaso-motor tone of the cutaneous vessels, the number and activity of the sweat glands and the covering of body hair, etc. (c) The efficiency of involuntary muscular exertions is different in different individuals; the work of circulation, respiration, vaso-motor control and peristalsis vary greatly with different persons. (d) The digestion is not a constant factor, individuals losing more or less carbon from the diet by alimentary fermentation, and this cannot be measured or controlled. (e) The work of the secretion of the digestive juices and the work of assimilation is different in different persons. All digestion is accompanied by excessive oxidation that is more marked for the digestion of protein than for that of fat and starch. This is variable, and this simply means that different persons accomplish the work of a unit of digestion more or less economically, just as different amounts of energy are required to carry different persons over a mile of running, depending on their economy of motion, which is an individual variation. (f) The oxygen input cannot be strictly controlled, some is lost in the intestinal gases, some from the skin, while some of the oxygen utilized in combustions comes from water and not from the atmosphere; therefore, an error is attached to all estimations of oxygen-input and carbon-dioxide-output. (g) The involuntary use of the voluntary muscles is different in different persons, an observation that is clearly noted in the phlegmatic and the nervous

This is unquestionably a factor of great influence. Now, sleep is an inconstant factor. Sleep is, however, on account of the muscular rest, attended by a saving in oxidation as against the waking hour. (h) The muscular habitus is of great influence. No two persons walk or run a mile with the same expenditure of energy. (i) The Calorie-kilo ratio will not be the same in the thin and fat, because of the thermic indifference of the fat, it be much lower in the obese. These numerous factors result in variations in the heat-unit per kilo per day ranging from 20 to 35 Calories. Following disease and during convalescence from emaciation, the figure is low. The findings in myxodema and following castration also fall within these factors. The relations in every case of obesity and in every case of emaciation (not due to disease) that has been studied by proper methods, with the above-named factors controlled or recognized, has led to negative results. All the properly studied cases have yielded results within the limits of the normal. No data exists, therefore, tending to show that there is a constitutional obesity or emaciation depending upon a retardation or acceleration of the carbonous metabolism. It is equally certain that no data exists tending to show that such a thing as a retardation of the protein metabolism occurs.

The incompletion of an act of nitrogenous or carbonous metabolism is known to occur under several circumstances. It is probable that future investigations will demonstrate that incompletions in the intermediary metabolism lie at the basis of many conditions now assumed to be intoxications solely on the basis of symptomatic analogy. The end-products of metabolism are quite free of toxic properties and we are thus compelled to revert to the intermediate metabolism for the chemical agents of systemic auto-intoxications. Several of these are known in the chemical sense, though in but one instance is a clinical entity connected therewith. If an excess of tyrosin be formed in the body, the liver will be unable to oxidize it all and a part of the substance will appear unchanged in the urine. In the alcaptonuric individual, however, a qualitative variation is observed in that the substance appears in the form of di-oxy-phenyl-acetic acid, which is normally an intermediary stage in the oxidation of tyrosin to di-oxyphenyl-a-oxy-proprionic acid. When the portal blood is deflected from the liver through an Eck fistula the ammonium salts, instead of being converted into urea, are eliminated in part unchanged. Idiopathic cystinuria presents another illustration of an incompleted act of protein metabolism, though here a new substance is produced. Normally we have the b-cystein in the body converted into taurin by the splitting off of one molecule of $CO_2$; in the cystinuria this reduction does not occur, but instead two molecules of the b-cystein are condensed to form cystin, which is eliminated in the urine. For the fat metabolism we are able to cite an instance of incompleted metabolism that is the cause of an intoxication. The normal urine contains a trace of acetone derived by reduction of diacetic acid, which is in turn derived by oxidation from b-oxy-butyric acid. An excess of fat in the diet is followed by an increase in the elimination of acetone. We may infer, therefore, that normally the oxidation of fat passes through the stage of b-oxy-butyric acid, of which a trace appears as acetone. In diabetes and other conditions, large quantities of diacetic and beta-oxybutyric acid appear in the urine, and the withdrawal of the body cations that results from the elimination of these acids is associated with the intoxication of diabetic coma. This is the only known auto-intoxication, of which the chemical agent has been demonstrated.

Instead of the normal end-products of metabolism, abnormal products might be formed. A probable illustration of this fact is seen in idiopathic ptomainuria. In certain individuals, some of them with idio-

pathic cystinuria, cadavarin and putrescin are regular constituents of the urine. Lysin and ornithin are normally formed in the course of metabolic protein hydrolysis (the ornithin being derived from the fermentative hydrolysis of arginine) and are finally converted into urea. It has been shown, however, that from the fermentation of lysin and ornithin, cadaverin and prutrescin may be derived, and in these cases they are in all probability so derived.

The hydrolysis of protein and the combustion of sugar and fat may under pathological conditions be increased. For the protein metabolism this may be interpreted to mean either an exaggeration of the normal autolysis, or the formation of an excess of the intercellular proteolytic ferments. For the carbonous metabolism it means that the normal law, that the combustion of these substances is related solely to the heat needs and the muscular exertions, is transgressed. This transgression may be attributed either to an excess of the glucolytic ferment, to the abnormal reversion of glycogen into sugar, or to the abnormal conversion of insoluble into soluble fat.

Fever per se tends to exaggerate the protein catabolism; the exaggeration is not marked, it may be absent. The febrile infections also tend to this result. This is a varying factor. Sometimes one sees the nitrogen balance undisturbed through a long febrile infection, if the powers of digestion have been good. In other instances the exaggeration of the protein catabolism is extreme. Usually in these infections the combustion of sugar and fat is much less exaggerated (it may be normal), so that the end result often is that the body of the convalescent is poorer in cellular protein than before illness, and relatively richer in fat. In these diseases the saving power of sugar for protein is lessened, just as in the minimal protein diet; one can reduce the nitrogen loss by the ingestion of large amounts of sugar or fat, but far more than the isodynamic amounts are necessary. In the convalescence from the infections in which the exaggeration of the protein catabolism has been marked, we observe one of the best-known instances of metabolic economy. Such a convalescent individual will retain nitrogen—that is, build cellular protoplasm—on a nitrogen input that would be scarcely sufficient in the same individual in health to maintain a balance. And such an individual will similarly lay on fat on a diet that normally would be scarcely enough to maintain his body heat. Under these circumstances every variable factor in the carbonous metabolism is operative in the direction of efficiency.

In exopthalmic goitre the protein catabolism may be much exaggerated, and forced feeding with protein may be unable to attain a balance. Whether this is a primary result of the thyroid disease, due to an excess of a hypothetical zymo-excitor, or whether it is secondary to the disturbed carbonous metabolism is not known. In diabetes, where a similar exaggeration is to be noted, the condition is quite certainly secondary to the disturbances in sugar combustion.

In pernicious anemia and leukemia, and during the progressive stages of malignant disease, rather marked exaggeration of the protein catabolism is observed. It is to be noted in many exogenous intoxications such as poisoning with phosphorus, chloroform and arsenic. For all of these it is probable that the exaggeration is due simply to an increase in the cellular autolysis of the body, and the disintegrated protoplasmic protein is, like any other excess of circulating protein, hydrolysed and burned.

An exaggeration of the carbonous metabolism probably occurs to some extent in most fevers. While disturbances in heat dissipation are the most potent factors in the production of fever, there is in most febrile conditions associated with bacterial infections some excess of heat formation. A very marked exaggeration in the combustions occurs in exopthalmic

goitre. In some of the well reported instances of this disease, the ingestion of over 60 Cal. per kilo per day (double the normal amount), was not sufficient to maintain the combustions without the loss of body fat. This is a positive exaggeration, due to a positive stimulation of the reactions of combustion, and not the result of an exaggerated heat dissipation. A marked exaggeration in the combustion of fat occurs in diabetes, as a compensation to the almost complete cessation in the combustion of sugar. A moderate excess of combustion has been observed in some case of pernicious anemia and leukemia, as well as in the cachexia of malignant disease. For most other diseases there have been no investigations.

It is apparently a rule that when either the carbonous or the nitrogenous catabolism is exaggerated, the reciprocal efficiency of the other is lowered, an excess of protein in the diet will not reduce the carbon combustions as in the normal, an excess of sugar or fat will not spare protein as in the normal. Just the opposite of this is to be observed during convalescence from disease (and in subnutrition), here the saving power of carbohydrate for protein and the reciprocal availability of protein for sugar and fat are increased.

## THE SPIROCHETA PALLIDA.*

### By SANFORD BLUM, M. D., San Francisco.

THE spirocheta pallida was first recognized by Schaudin in a smear preparation obtained from a secondary syphilitic papule in March, 1905. The preparation was examined fresh and the spirocheta was first observed in its motile state. Subsequently Schaudin and Hoffmann succeeded in staining the organism and studied it also in the colored preparation. Since then various investigators have studied this interesting organism and valuable information has been contributed by Lowenthal, Metchinkoff and Roux, Buschka and Fisher and others.

Up to the present time the spirocheta pallida has been found in syphilitic subjects in the discharge from eroded primary and secondary lesions, in the deep unexposed portions of primary and secondary lesions, in smears obtained from extirpated luetic inguinal glands and from the sap obtained from luetic inguinal glands by aspiration, in deep tissue smears from isolated intact papules, in the fluid contained in pemphigus vescicles in a child congenitally luetic, in smears from the liver and spleen of a child dying from hereditary lues, and perhaps in the blood taken from the finger of a luetic individual. It has not been found except in luetic cases.

Morphologically the spirocheta pallida is a small, delicate, corkscrew-like spiral with tapering ends. It varies in length from 3 mu to 14 mu and its thickness may be estimated at ¼ mu. Each spirocheta has from 3 to 14 twists or coils. It is extremely faintly refractive and takes stains with difficulty. In life it moves rapidly by a rotary motion in the direction of its long axis. First it moves in one direction; then comes to a standstill and then moves in the opposite direction. Schaudin describes other movements which it executes without locomotion, which he ascribes to an expression of the play of an undulating membrane. These are undulatory motions involving the entire structure.

In life the spirocheta pallida differs from other spirocheta in its smallness, delicacy and much fainter refraction of light, and especially in the nature of its coils. These can best be described as corkscrew-like. They are regular, narrow and deeply bent and these characteristics are constant, whatever the source of the individual specimen may be (primary lesion, gland, papule, monkey, etc.). They have the same configuration in the stained as in

*A resume of investigations to date, with demonstration of specimen, before the California Academy of Medicine, August, 1905.

fresh preparations and it may be added that they stain faintly. They do not take most of the ordinary stains and when they stain they do so more faintly than the other spirochetæ.

The technic of staining as given by Hoffman is as follows:

(1) Make a thin smear and dry in the air; (2) fix the preparation in absolute alcohol (preferably without delay); (3) dry, and stain with Giemsa's mixture. (Hoffmann uses Grübler's preparation. Of this 10-15 drops are added to 10cc distilled water. In this the preparation is stained for an hour in a porcelain dish.); (4) dry in the air and affix to slide with cedar oil or Canada balsam.

Gonder and Hoffmann succeeded in staining with fuchsin and Anilin water gentian violet.

Schaudin believes that the spirocheta family differs from the spirillum family in that the spirocheta are protozoa, while the spirilla are bacteria. He differentiates the spirocheta pallida from the other spirocheta—sp. refringens; sp. balanoposthitis; sp.
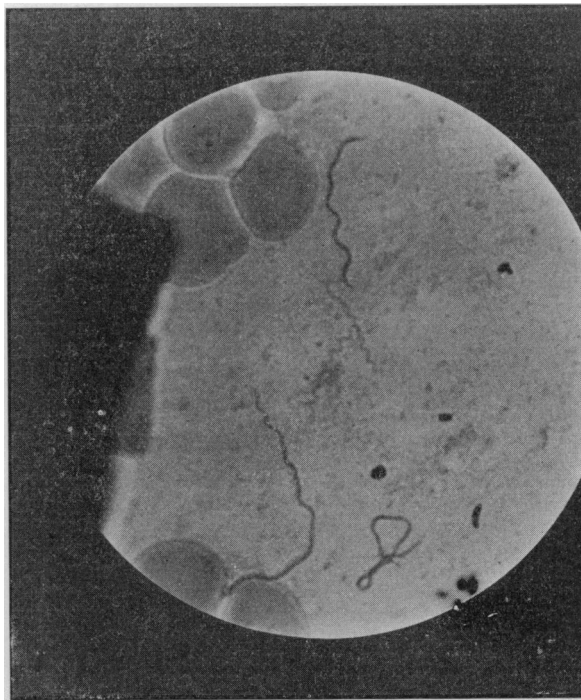


Microphotograph showing the delicate spirocheta pallida and also the coarser spirocheta refringens.

erosiva circinata; sp. buccalis; sp. angina Vincenti; Anserina, Chicken, and Obermeieri—by their smallness, their greater delicacy, the nature of their spirals, their much feebler refractibility and the difficulty with which they take stains.

The cases in which Schaudin and Hoffmann found the spirocheta pallida were recent cases—4½ to 5 months old. In one instance Hoffmann found the spirocheta pallida in the blood obtained by puncturing the spleen of a man with recent lues a day before the roseola appeared. Drs. Buschka and Fisher found it in the spleen of a child dying from hereditary lues. It has also been reported as found in blood taken from the finger of a luetic individual. Schaudin found it in the pemphigus vesicles, liver and spleeen of a child afflicted with hereditary syphilis. Metchnikoff and Roux found it in the primary lesions of inoculated monkeys.

The work of Lowenthal is important. He found 9 intracellular spirocheta in one cell in the deep tissues